CARDIAC DYSRHYTHM MECHANISMS AND ANALYSIS OF AFTERDEPOLARIZATIONS: TRIGGERED ACTIVITY

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Based in Frank Starmer hypotheses*

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Cardiac arrhythmias or dysrhtymias is used for any alteration in formation (automaticity) and/or in the conduction of the electrical stimulus (dromotropism) in the heart. (1). We should add to this concept, those generated by triggered activity originated by oscillations in action potential in phase s 2 or 3 named Early After Depolarization (EAD) and in late in phase 4 (Delayed After Depolarization: DAD). Note: triggered activity is a variety of the sustained activity originated by post-depolarizations that depend on the preceding action potential, and consequently, it is not automatic. (2).

An arrhythmia is an abnormality in the rhythm or the pattern of heartbeats, which may be slow (bradyarrhythmia), fast (tachyarrhythmia), premature (extrasystole or premature contractions), delayed non-sustained (escape), delayed sustained (substitution) or irregular (e.g., fibrillation). Note: the term dysrhythmia is more appropriate than arrhythmia, because it means altered rhythm and not loss or absence of rhythm, as the word arrhythmia suggests. (3). Both terms are used in literature as if interchangeable.

Table 1 shows the different mechanism of cardiac arrhythmias

Table 1

CLASSIFICATIONS OF CARDIAC DYSRHYTHMIAS BY THEIR MECHANISM

1. Dromotropic or by reentry

- Anatomically determined reentry: It may be found in the atria, as in the case of paroxysmal atrial tachycardia secondary to atrial micro-reentry (intra-atrial reentry), known as reentrant atrial tachycardia or tachycardia secondary to incision, by the presence in the atria of a reentry circuit located in the atria around a region of intra-atrial suturing with two pathways of different conduction velocities and refractory periods. Characteristics of micro-reentry secondary to atrial incisionare: It may be terminated by programmed atrial pacing (useful to differentiate it from the automatic one); positive “entrainment” that confirms the reentry mechanism; paroxysmal character: sudden onset and end; atrial rate between 130 bpm and 150 bpm (less than the automatic one); absence of irregularity in the initial phase of the event or warm-up period; first P’ beat of morphology different from the rest of the tachycardia; Variable duration: seconds (transitory), minutes, hours (sustained: the most frequent one), months or years (persistent or incessant); frequently confused with atrial flutter; observed in the cases of mega-atria by COPD. In 25% of the cases of correction of complex congenital heart diseases (ASD correction, post-surgery of great vessel transposition, such as the surgery of Mustard, Sennig or Fontan) or cardiomyopathies; refractory to drugs and with a high degree of response to the procedure of ablation by radiofrequency.

- **Functional reentry leading circle and “phase 2 reentry”:** Another way of dromotropic mechanism in the genesis of arrhythmias, is the so-called functional or free reentry, which is processed in absence of a defined pathway or circuit, or anatomical structure, which occurs in the neighboring fibers that have different electrophysiological properties, with local

differences in AP characteristics, based on rapid and slow longitudinal conduction properties, and thus the stimulus advances following the trajectory where there is the least refractoriness. This variety was called leading circle (4). In this animal model in rats, the length of the pathway is not defined by an anatomic obstacle, but it is by the electrophysiological properties of the myocardium. A dispersion of excitability and/or refractoriness, as well as the anisotropic distribution of intercellular resistances, condition its installation and perpetuation. As the arrhythmias secondary to this mechanism are due to variable pathways, and they are not anatomically defined, they usually present polymorphic morphology. The central portion of the free reentry is known as core. Atrial fibrillation is an example of leading circle reentry type, in which a reentrant circuit spreads from a refractory core according to its trajectory, by the fibers that possess the shortest refractory period, being blocked in the fibers with more prolonged refractory periods. The conversion from free or functional reentry into anatomical reentry is called pinning or anchoring. The pathogenesis of Brugada syndrome is phase 2 reentry resulting from shortening of the epicardial action potential duration at the right ventricular outflow tract (RVOT). (5).

- **Anisotropic reentry** Anisotropy, the property of being directionally dependent, is ubiquitous in nature. Propagation of the electrical impulse in cardiac tissue is anisotropic, a property that is determined by molecular, cellular, and histological determinants. The properties and spatial arrangement of connexin molecules, the cell size and geometry, and the fiber orientation and arrangement are examples of structural determinants of anisotropy. Anisotropy is not a static property but is subject to dynamic functional regulation, mediated by modulation of gap junctional conductance. Tissue repolarization is also anisotropic. The relevance of anisotropy extends beyond normal propagation and has important implications in pathological states, as a

potential substrate for abnormal rhythms and reentry. (6). It originates from a greater conduction velocity in the longitudinal direction of the fiber, and slower transversally or perpendicularly. This velocity difference favoring the longitudinal direction, is a consequence of the greater density of the so-called “gap-junctions” in the ends of the cells in comparison to the lateral area. The desmosome, specialized area of the membrane, presents an area of intercalated discs, which hold the cohesion between two cells, called “gap junction” or nexus responsible for the syncytial character of the cardiac muscle, which determines the “all or nothing” type of response of the cells. Such response consists of reaching the limit of a stimulus and then, obtaining a response. Although we may increase the intensity of the former, the response will always display the same intensity.

- **Reflection:** The impulse in superior part reaches an area severely involved in its dromotropic ability, and is blocked. In the inferior part, it slowly advances to go back through the opposite direction by the reflection mechanism (reflection) and later be conduced slowly, and reenter toward its origin (7). Propensity for atrial fibrillation (AF) in patients with an accessory pathways(AP) is strongly related to preexcitation, larger atria, male gender, and older age. Reflection and microreentry at the AP may be important for AF initiation in patients with manifest (preexcited) Wolff-Parkinson-White syndrome. Similar mechanisms also may trigger AF in patients without an AP.(8).

- **Summation:** Two independent impulses (A and B) when converging jointly in a decremental conduction area (C), are joined and manage to emerge later from such area (D). The passage of the stimulus in the decremental area would not be possible for either of them in isolation.

2. Automaticity

- **Enhanced normal automaticity or hyper-automaticity** arrhythmias generated by these mechanisms occur in any site of the heart where there are fast fibers (more negative rest potentials): Purkinje fibers and even contractile atrial and ventricular myocardium. This type of arrhythmia depends on the rapid Na+ channel and therefore, it is suppressed by overdrive suppression, a distinctive characteristic of normal automatic mechanisms.The following are causes of enhanced normal automaticity: 1) Beta-adrenergic stimulus: it increases the ascending ramp in phase 4 (it becomes steeper)(9); 2) Hypopotassemia: extracellular K+ between 3 and 4 mEq/L depresses the ramp;(10); 3) Mechanic distension;4) Ischemia. The main arrhythmias by enhanced normal automaticity or hyperautomaticity are: inappropriate, permanent or non-paroxysmal sinus tachycardia, atrial tachycardia, junctional tachycardia and Accelerated IdioVentricular Rhythm (AIVR).
- **Abnormal automaticity.** when action potential is originated spontaneously, and when the rest potential reaches values more negative than –70 mV. A basic condition for this mechanism is that the membrane spontaneously reaches levels lower than –70 mV (very reduced membrane potential). Abnormal automaticity is not abolished by overdrive suppression. A reduction in maximum diastolic potential (MDP) is observed, which corresponds to the end of phase 3 and the onset of phase 4, and is always dependent on the slow Ca2+ channel. It may occur in contractile non-automatic cells, when the membrane potential reaches –60 mV. The main causes fo arrhtymias secondary to abnormal automaticity are: Atrial tachycardia excluding the one caused by digitalis intoxication, AIVR (Accelerated IdioVentricular Rhythm), junctional tachycardia, VT during the first five days after acute infarction and any cause that increases maximum diastolic potential (MDP) depth decreases automaticity. E.g.: acetylcholine.

3. By triggered activity or after-depolarizations

- **Early After Depolarization(EAD)**
- **Delayed After Depolarization(DAD)**

4. Mixed

In this manuscript we devoted to arrhythmias consequence of triggered activity or after-depolarization.

Afterdepolarizations are abnormal depolarizations of cardiac myocytes that interrupt phase 2, phase 3, or phase 4 of the cardiac action potential (AP) in the electrical conduction system of the heart. Afterdepolarizations may lead to cardiac arrhythmias.

Early afterdepolarizations

Early afterdepolarizations (EADs) occur with abnormal depolarization during phase 2 or phase 3, and are caused by an increase in the frequency of abortive APs before normal repolarization is completed. Phase 2, dome or plateau may be interrupted due to augmented opening of calcium channels.

They are divided into:

I) PHASE 2 EADs

- Oscillations that occur when in the plateau, dome or phase 2 by increase in inward Ca2+ by the slow *I*Ca-Lchannel.
- There is an additional and persistent inflow of the sodium cation in phase 2 or AP plateau. This is observed in variant 3 of long QT syndrome or LQT3. This explains the increase in ST segment duration in ECG. QT interval prolongation at the expense of ST segment prolongation with delayed T wave onset.

II) PHASE 3 EADs

- Phase 3 interruptions are due to the opening of sodium channels. EADs can result in torsades de pointes (TdP), tachycardia, and other arrhythmias. These post-depolarizations occur during phase 3 of AP by reduction in the activity of outward K+ channels (Ik-R or Ik-s) as it happens in congenital long QT syndromes LQT2 and LQT1 respectively. The latter differentiate from the former in that they present Ca2+ release from the Ca2+ release channel or ryanodine receptor. Moreover, activation of the INa+-Ca2+ cation exchange channel by electrogenic mechanism (there is exchange of three Na+ molecules by one of Ca2+. Afterhyperpolarizations can also occur in cortical pyramidal neurons(11). There, they typically follow an AP and are mediated by voltage gated

sodium or chloride channels. This phenomenon requires potassium channels to close quickly to limit repolarization. It is responsible for the difference between regular spiking and intrinsically bursting pyramidal neurons.

CLINICAL CAUSES OF EADs

1. Drugs: Sotalol, NAPA, quinidine, pshychotropic agents (phenothiazide), phenothiazide, anthopleurin A1, Bay K 8644, (12).
2. Ketanserin a 5-HT2 antagonist, directly inhibits the ATP-sensitive potassium channel
3. Hypoxia.
4. Hypothermia.
5. Acidosis.
6. Aconitine.
7. Ethylenediaminetetraacetic acid (EDTA).
8. Effect of Cesium.
9. Hyperadrenergic state. E.g.: Subarachnoid hemorrhage.
10. Mitral valve prolapse

Note: EADs originate Torsades de Pointes.

To develop a better understanding of the physical mechanism that determines transitions between normal monotonic repolarization and spontaneous oscillation it is necessary to know the mechanism behind spontaneous oscillation in cardiac cells and EADs in particular is sort of fuzzy. While it is well known that reopening of inward channels during repolarization is required to establish a net inward current (and hence initiate a transition from repolarization to depolarization), the details are still fuzzy. From a qualitative point of view, reopening Ca2+ or Na+ channels are required for one or more EADs and many investigators are satisfied with this level of detail. But for others, under what circumstances will reopening channels lead to EADs? Channels open and close

all the time - under many different conditions. So what is special about repolarization that leads from channel reopenings to EADs? . From a physical mechanism perspective - under what conditions will reopening inward channels lead to EADs? Investigations revealed three different conditions where the role of reopening could be explored:

1. Reopening during the positive resistance region of the repolarizing i/v curve
2. Reopening during the negative resistance region of the repolarizing i/v curve; and
3. Reopening when the membrane potential is precisely at the transition from the positive to the negative resistance region.

While these three conditions are clear to a few (most notably, Otto Hauswirth, Denis Noble and Dick Tsien), there is almost nothing in the literature linking these ideas to EADs. Measuring the propensity of EADs during repolarization by measuring the development of "the EAD safety factor" as a function of s1-s2 delay. *The safety factor is the current amplitude of the transition from + to - resistance during repolarization*. It all started in 1988 when a patient was admitted to the Duke ER with a ventricular tachyarrhythmia - apparently secondary to an overdose of propolxyphene (use-dependent Na+ channel blocker). How such arrhythmias might be triggered?. This led to some insights into how use-dependent Na+ channel blockade increases the duration of the cardiac vulnerable period (1991). This was followed (1995) by studies of spiral waves and how reducing K currents can force a transition from circular spiral tip movement (monomorphic reentry) to meandering (polymorphic reentry) - sort of like TdP. Still how reentrant arrhythmias were triggered? Some ideas seemed to spontaneously come together - 10 years being fermented - Most of the numerical experiments use Sergey Missan's CESE system, a java-based numerical electrophysiology experiment environment.

Background

Following initial depolarization, cardiac cells either monotonically repolarize and return to the normal rest potential or exhibit EADs (plateau oscillations) followed either by completion of repolarization or incompletely repolarize and equilibrate

at a depolarized equilibrium potential. Depending on their gating voltage and time dependence, previously inactivated Na+ and Ca2+ channels return to the open state and reduce the net repolarizing current. As long as the repolarizing current is outward, repolarization continues.

Early studies of Hauswirth, Noble and Tsien (Hauswirth, O., Noble, D. and Tsien, R.W. The mechanism of oscillatory activity at low membrane potentials in cardiac Purkinje fibres J. Physiol. 1969 200:255-265) revealed that oscillations at plateau potentials in cardiac Purkinje fibers were associated with the dynamics of the delayed rectifier and a source of inward current, now recognized to be supplied by reopening Ca2+ channels ((13;14;15;16). Moreover they identified the negative resistance region of the repolarizing i/v to be critical in establishing oscillatory activity.

Detailed exploration of the cellular EAD mechanism using in vitro studies has been hindered by the absence of a method to precisely control the time and voltage dependent conductances as they change during repolarization. On the other hand, numerical studies provide the control necessary to dissect many of the complexities of membrane repolarization and demonstrate associations between cellular processes. Such studies have provided important insights linking a number of different modulators of inward and outward currents to EAD production (17;18).

Mutant channels, ion channel blockers, exercise, stress, sleep, drugs that alter transmembrane ion transporters and internal calcium stores, as well as heart rate and pause duration have been associated with conditions associated with sudden cardiac death, supposedly triggered by EADs (19;20;21;22;23;24) However, it is still unclear how or whether these observations can be framed in a simple mechanistic characterization of the transition from normal repolarization to oscillatory repolarization. The transition from monotonic repolarization to oscillations results from the coincidence of the repolarizing membrane potential and the minimum of the current-voltage relationship at the exact moment the minimum of the negative resistance region = 0. Consequently, proarrhythmia is amplified during repolarization by any increase in availability of inward currents (e.g. delayed inactivation of the fast Na channel) and/or any decrease in the availability of outward repolarizing currents

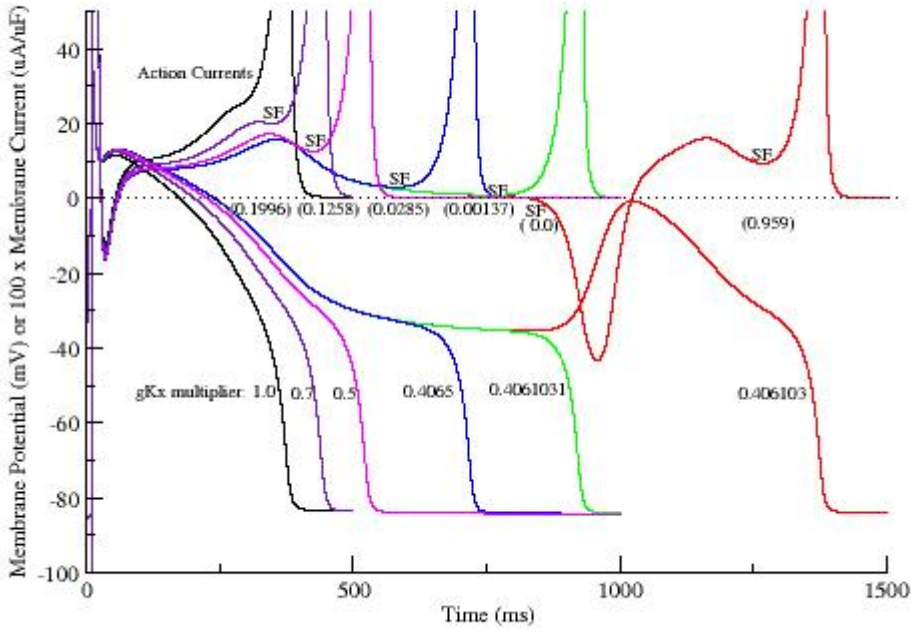
(e.g. loss-of-function in HERG channels). These results also suggest that APD prolongation with a squared repolarization morphology will minimize the time available to develop the negative resistance region and thus be more antiarrhythmic than APD prolongation with triangular-like repolarization.

The Problem and Insights

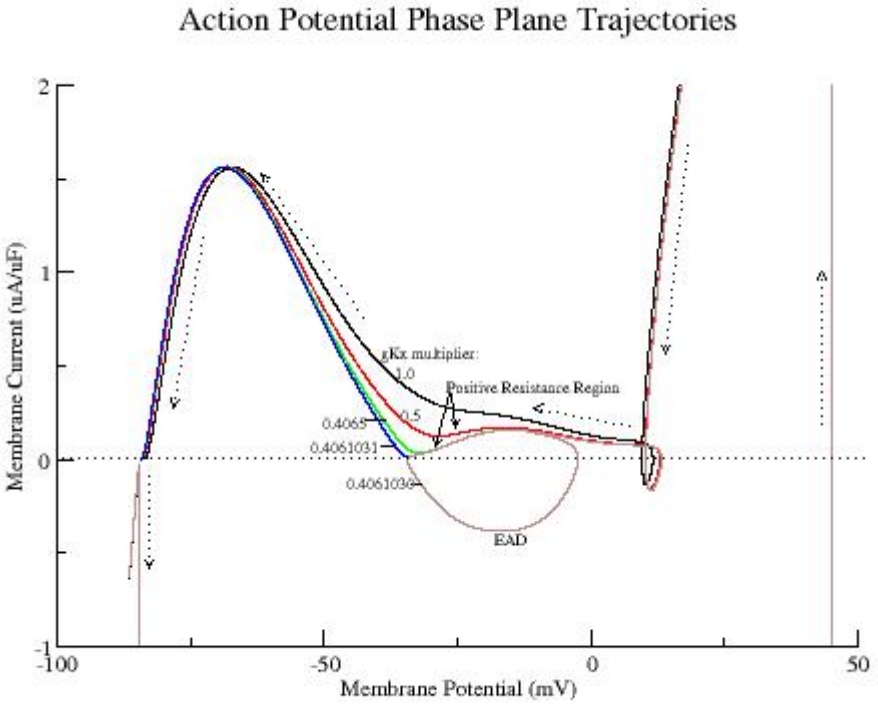
One way to explore EADs is to trigger APs with progressively smaller repolarizing currents and observe the relationship between the membrane current and membrane voltage. Shown here are the APs and their action currents where the delayed rectifier conductance was progressively reduced, thereby slowing repolarization and increasing the APD. As repolarization slowed, the APD increased and a positive resistance region developed (a result of increasing inward currrents) where reducing Vm reduced the membrane current (indicated by SF) and produced a plateau in the AP. The transition from monotonic repolarization to EADs did not occur until two conditions were met:

- The minimum of the transition from positive resistance to negative resistance region (SF) was 0 uA/uF and
- The membrane reached the potential associated with SF=0 at the same time the SF became zero.

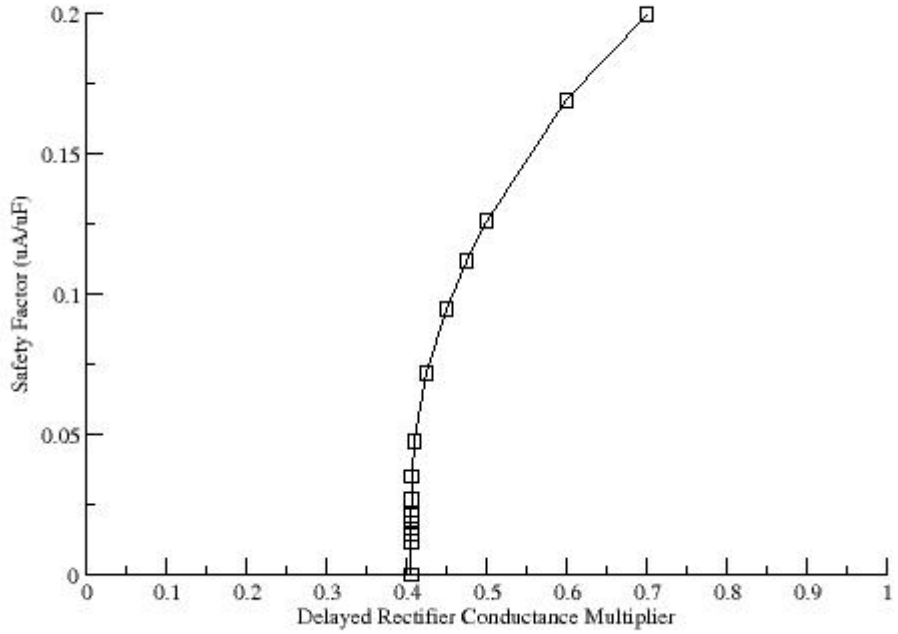
The transition from monotonic repolarization to oscillation (EAD) occured at gKx = 0.4061030*gKx(max) using the LR1 model.



The phase plane portrait illustrates the two competing processes - that of membrane repolarization and that of growth of the negative resistance region. Here is shown phase portraits of a control AP, two intermediate APs (gK multiplier = 0.5, 0.4065), an AP near the transition from monotonic repolarization to EADs (gK multiplier = 0.4061031) and an AP with an EAD (gK multiplier = 0.4061030). Slowing repolarization gradually reduced the minimum of the negative resistance region (Safety Factor, SF). Note the transition from monotonic repolarization to EAD occurred when the phase point (Vm, Im) touched the i=0 axis.

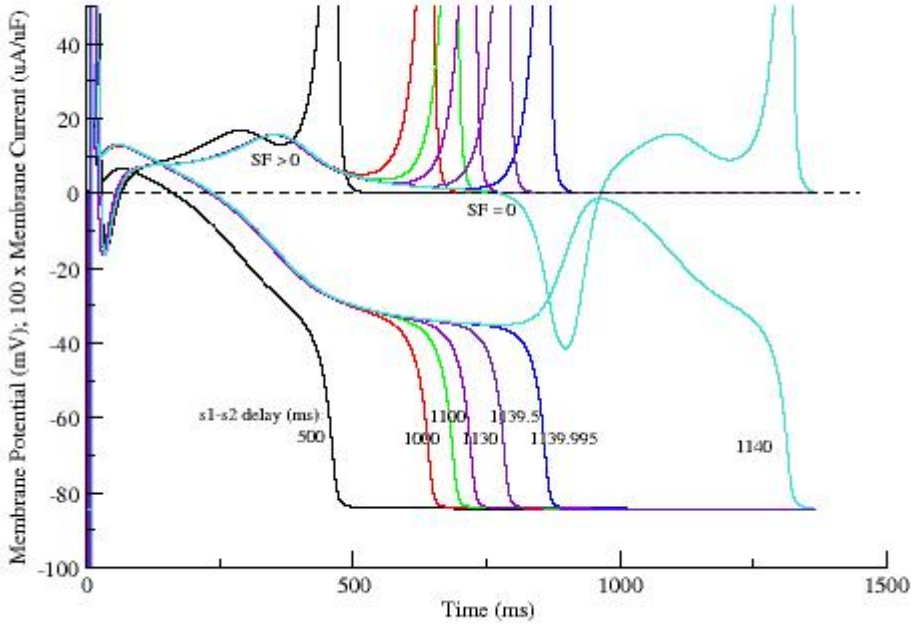


For the LR1 ventricular model, the control AP shows and outward negative resistance region until a transition back to a positive resistance region at about -60 mV. As repolarization is slowed secondary to reducing gKx, a small outward positive resistance region develops at about -20 mV, passes through a minimum to a negative resistance region and then returns to a positive resistance at about -60 mV. For multipliers > 0.7 there is no negative-positive-negative transition in resistance so that there is no minimum and no SF. Shown below is the value of the minimum (SF) as a function of gKx.

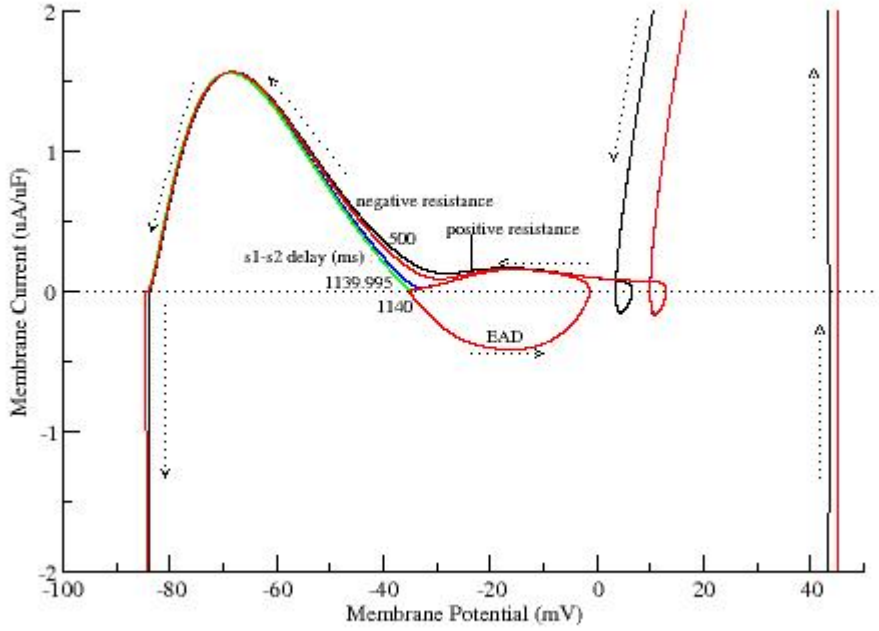


Many cellular processes can modulate the amplitude of inward and outward currents. Specifically a regular rhythm results in accumulated open delayed rectifier channels as well as alterations in the Ca^{2+} stores. If the SF hypothesis is correct, then SF profiles measured under dynamic conditions using a conditioning pulse train followed by a delay (s1-s2) and a test pulse should display the similar SF reductions secondary to deactivated IK during repolarization as s1-s2 delay increases observed with SF profiles secondary to explicit reduction in gKx measured from APs initiated from equilibrium initial conditions.

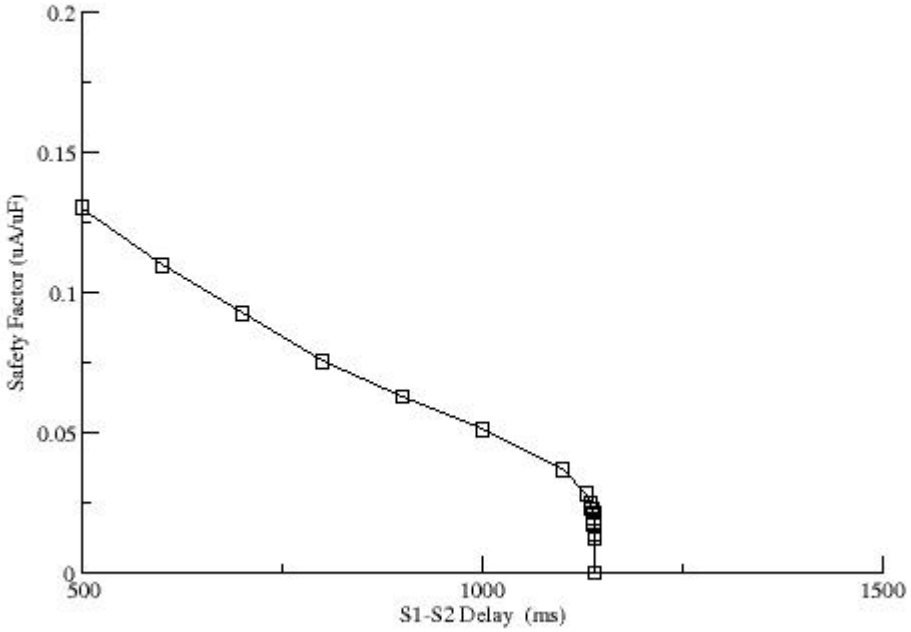
To demonstrate the generic property of the SF mechanism, plotted the APs and action currents as a function of s1-s2 delay following 40 conditioning pulses at 2Hz. Note the monotonic reduction in SF as the s1-s2 delay is extended. The earliest AP (black) was with s1-s2 = 500 ms, the same as the conditiong 2Hz train. The EAD occurred when s1-s2 = 1140 ms. Shown also are intermediate are APs and action currents with delays ranging from 1000 ms to 1139.995 ms.



Shown here is the increase of the positive resistance region as a function of s1-s2 delay



Similar to explicitly reducing gKx, implicit reduction of gKx by increasing the s1-s2 delay reduces the SF.



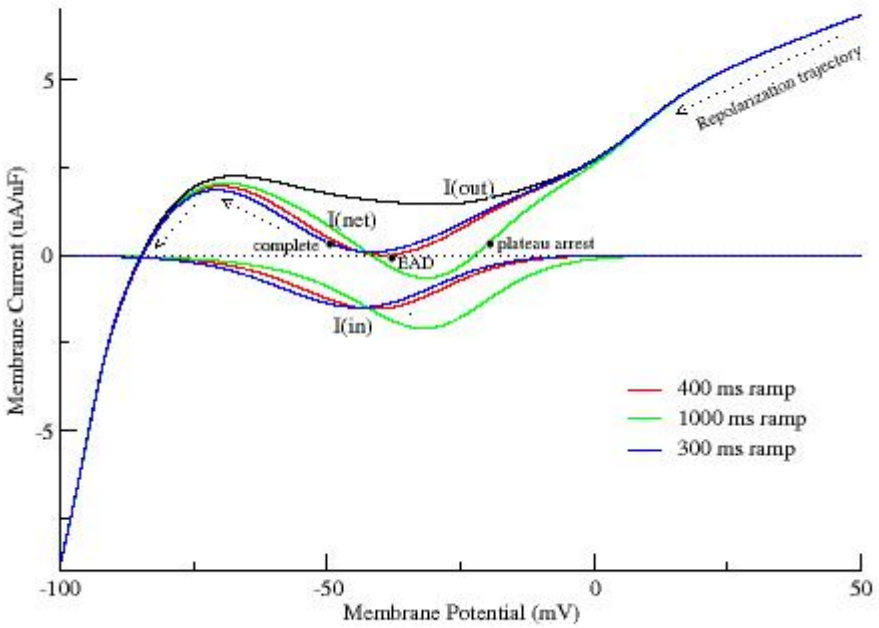
A Qualitative Model

These observations demonstrate that repolarization morphology is sensitive to the location of the repolarizing membrane potential, Vm, relative to the negative resistance region of the repolarizing i/v. Specifically, if the membrane potential enters the negative resistance region of the repolarizing i/v before the SF = 0, then repolarization accelerates (phase 3) and terminates after reaching the normal rest potential. If, on the other hand, the membrane potential remains on the positive resistance limb of the repolarizing i/v after the SF = 0, then repolarization is stabilized with the result of plateau arrest. Finally, if the membrane potential reaches the minimum of the negative resistance region at the moment that the SF = 0, then an EAD will result. When Vm reaches the minimum and SF = 0, repolarization is transiently arrested (because the net membrane current is zero) and as the Ca channels continue to recover, the SF becomes negative and Vm gradually shifts from repolarization to depolarization.

Below are plotted three i/v curves using a reverse ramp from +50 to -100 mV with durations of 300, 400 and 1000 ms. Shown is the total outward current,

I(out), the total inward current, I(in), and the net transmembrane current, I(net), = I(in) + I(out). While the total outward current was insensitive to repolarization rate, the total inward current increased as the repolarization rate slowed and the minimum of the negative resistance region shifted to more depolarized potentials. The dots near the minimum of the negative resistance region represent the three possible relationships between Vm and the minimum of the i/v curve. If Vm passes through the mimimum of the i/v and into the negative resistance region before SF = 0 (blue curve), then repolarization continues. If Vm does not reach the minimum of the i/v before the SF = 0 (green curve), then as repolarization continues Ca channels will continue to reopen, shifting the minimum below the i=0 axis and resulting in a stable equilibrium and plateau arrest. However, if the membrane potential arrives at the minimum of the negative resistance region (red curve) at the same time that SF = 0, then an EAD will result. The coincidence temporarily arrests repolarization while Ca channels continue to open, thus initiating a depolarization.

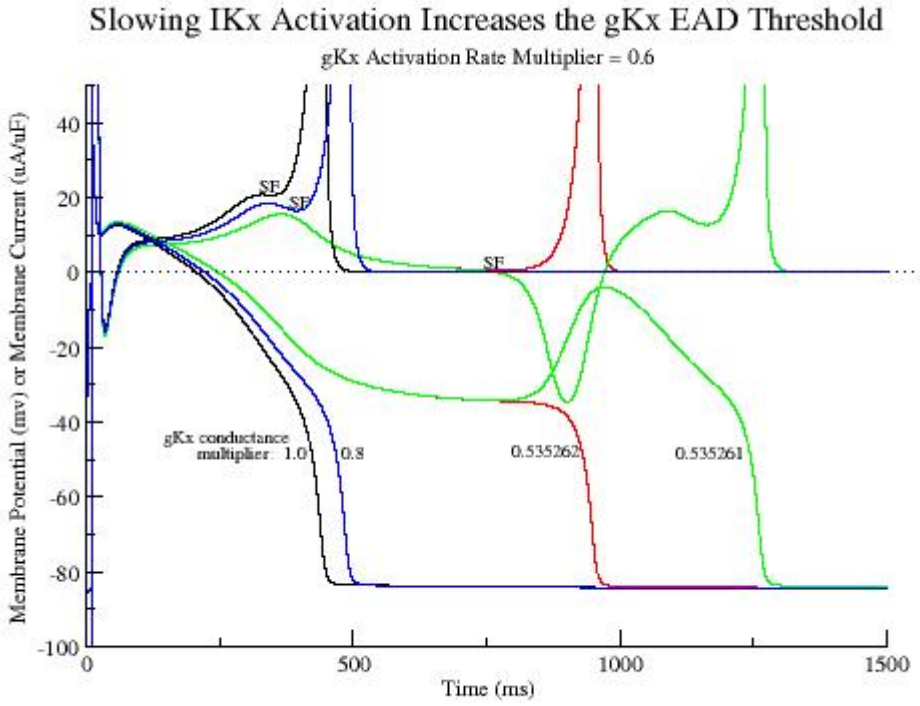
Why does coincidence of Vm and the minimum of the negative resistance region at the moment that SF = 0 lead to an EAD? As shown below, as repolarization is slowed, the peak of the inward current increases and shifts to more depolarized potentials. At the moment that I(Vm) = 0, repolarization is arrested but Ca channels continue to open and increase the total inward current. Thus the instantaneous current at the phase point becomes negative and depolarization is initiated, but at a very slow rate since the total current is very small. As slow depolarization increases, I(Ca) continues to increase thus accelerating the rate of depolarization. Only after Vm reaches the minimum of the I(Ca) current voltage relationship does the rate of repolarization slow and approach the point where I(net) = 0 where the current becomes outward and Vm returns to a repolarizing trajectory as shown in the above phase plot.



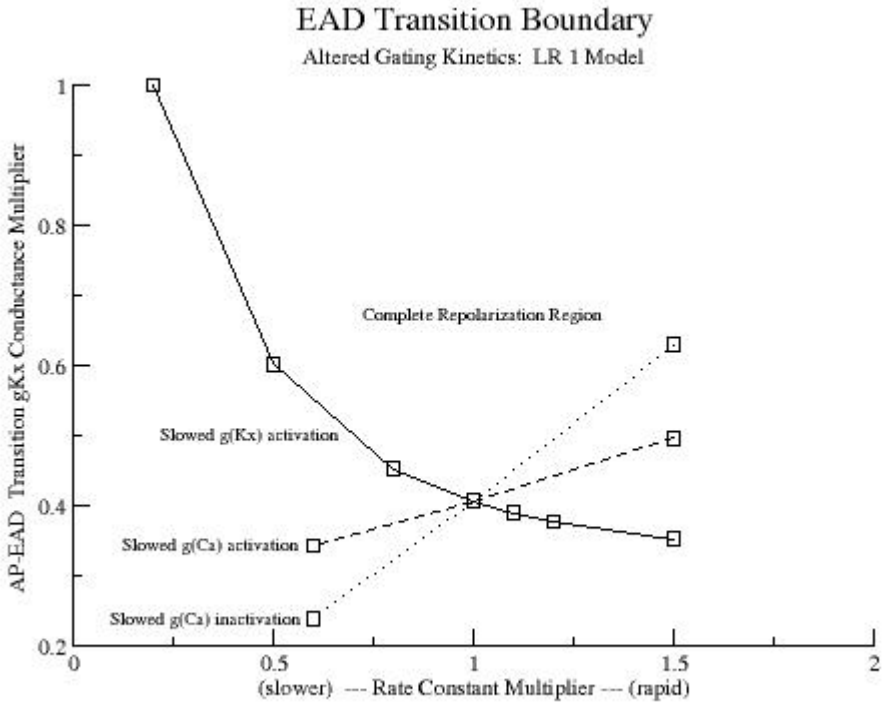
EAD sensitivity to altered gating kinetics

Perhaps most interesting is how altered gating kinetics change the window of EAD susceptibility. Mutant HERG channels not only have a reduced conductance but the activation kinetics is slowed. Consequently, based on the above model, we would expect slowed activation kinetics to extend the EAD susceptibility window by further slowing repolarization beyond that associated with reducing gKr. Similarly, increasing the activation rate of Ca channels should amplify the EAD susceptibility range by increasing the rate at which the secondary positive resistance region develops. Below are show the transition (EAD - complete repolarization) conductance multiplier as a function of the activation time constant. Rate multipliers less than 1 reflect slowed activation while multipliers greater than 1 reflect accelerated activation. (By rate multipliers I use multiplier*alpha and multiplier*beta and for the conductance multiplier, I use multiplier * gKx.

Slowing the activation time rate by 0.6 increases the threshold for AP - EAD transition from 0.4061031 to 0.535262 - suggesting that slower kinetics result in EADs with less loss of function (reduced macroscopic conductance)



As the IKx activation rate is slowed, the AP-EAD gKx transition conductance dramatically increases. Similarly, as ICa activation is slowed, the AP-EAD gIx transition conductance is diminished



These results suggest that mutations resulting in loss of function by reducing GKr or GKs increase the likelihood of EADs by increasing the likelihood that the phase point of the AP will find itself within the transition window. Further, mutations that increase the time constant of gating changes increase the vulnerability to EADs by increasing the range of GKr or GKs that will place the AP phase point within the transition region.

CHARACTERIZATION OF EADs

1) Greater incidence during low heart rates (bradycardia-dependent);
2) They occur faced with AP duration prolongation;
3) They end when repolarization is complete;
4) They are suppressed by “fast pacing”
5) They are observed in two AP levels: between 0 and –30 mV and between –60 mV and –70 mV.
6) Tendency to occur in runs.

Sort of a summary

For EADs, the critical issue is one of the comparative timing of the repolarization potential and the location of the minimum of the transition from positive to negative resistance. If they coincide when the current associated with the minimum is zero, then EADs will occur. A safety factor (SF) as the current associated with the minimum of the transition from positive to negative resistance. Now to better understand the influence of timing on EAD production, consider the following three options:

- Vm passes the minimum of the resistance transition region while SF > 0 This condition always results in accelerated repolarization since the membrane potential, Vm, is on the negative resistance limb of the iv when the current is outward.
- The minimum of the resistance transition region passes through i=0 before Vm gets there. The result is either sustained depolarization or a long long wait until intracellular Ca stores force repolarization.

- Vm reaches the minimum of the resistance transition region at exactly the same time as SF = 0. This results in an EAD.

Now complete repolarization is easy. If Vm manages enter the negative resistance region before SF=0, then repolarization is accelerated due to the negative resistance. One could say, but what if Vm was just barely past the minimum, could not continued restoration of gCa pull Vm down and start an EAD? The answer is no - at least from these numerical studies. For this to happen, there would be an increase in repolarization rate (as Vm enters the negative resistance region) before an EAD is triggered. I have never observed this in any numerical studies and think there must also be an mechanistic answer to the question.

Now what about Vm is near the minimum but on the positive resistance side of the i/v. In this case EADs are not possible because if Vm continued to repolarize, but the instantaneous current became negative, then the inward current would drive it back to a depolarized potential where i=0 - a stable equilibrium. In this case, there would be sustained depolarization (at least until the intracellular calcium stores and Na/Ca exchange did their thing while Vm hung around at the sustained depolarization potential and forced continuation of repolarization). This is not seen in either BR or LR1, but is easily seen with LR2 with its more accurate Ca model.

So that leaves Vm arriving at the minimum of the i/v at precisely the same time that the SF = 0. Continued recover of gCa will move the minimum into inward current territory and once Vm crosses over i=0 (in the phase plane), the EAD oscillation is triggered.

There are several predictions based on this model and logic

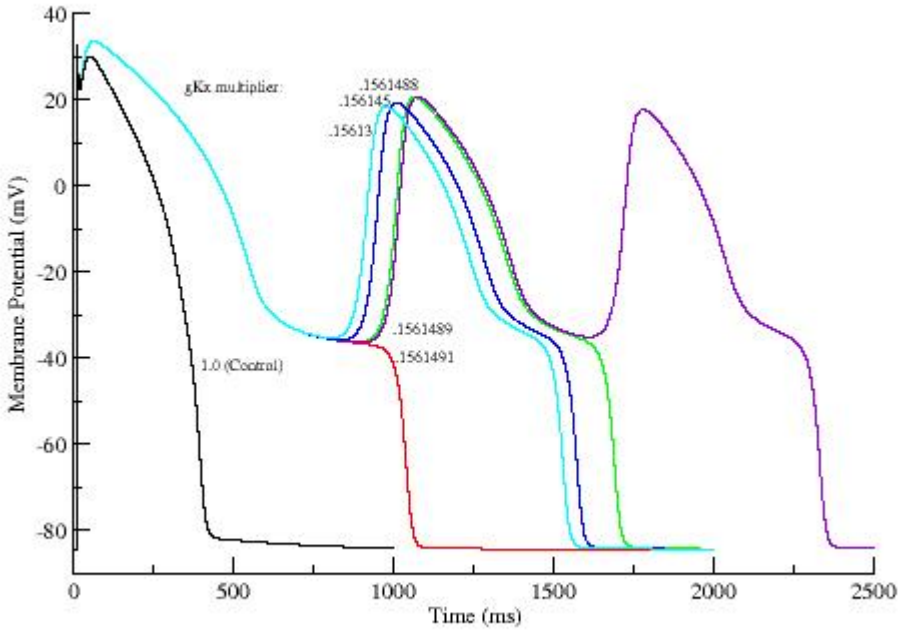
- Higher repolarization rates will be associated with longer delays before EAD takeoff
- Higher repolarization rates will be associated with more negative takeoff potentials.

Below are APs obtained from the Beeler Reuter, LR1 and LR2 model. Values of the delayed rectifier (BR and LR1) and (IKr, IKs for LR2) reduced and APs generated. Shown are control APs, an AP at the just before the transition to

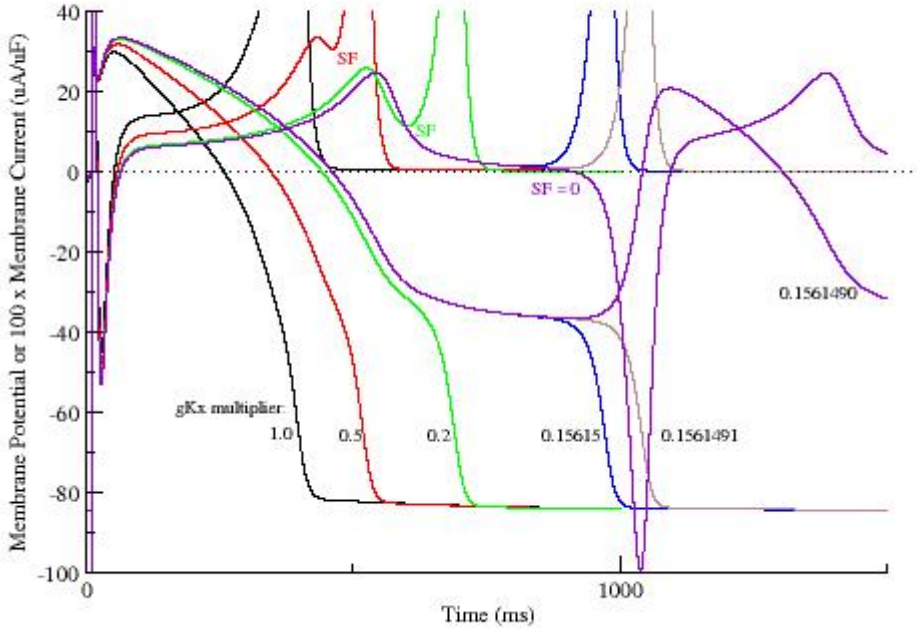
EADs and a group of APs associated with continued reduction in gK. Note that in all three cases, the maximum delay to takeoff was associated with the fastest overall repolarization (larger value of gK). As gK was reduced, the takeoff time was similar reduced and the takeoff potential was more depolarized. These results are consistent with the above hypothesis.

Demonstration of comparable results from BR, LR1 and LRd models

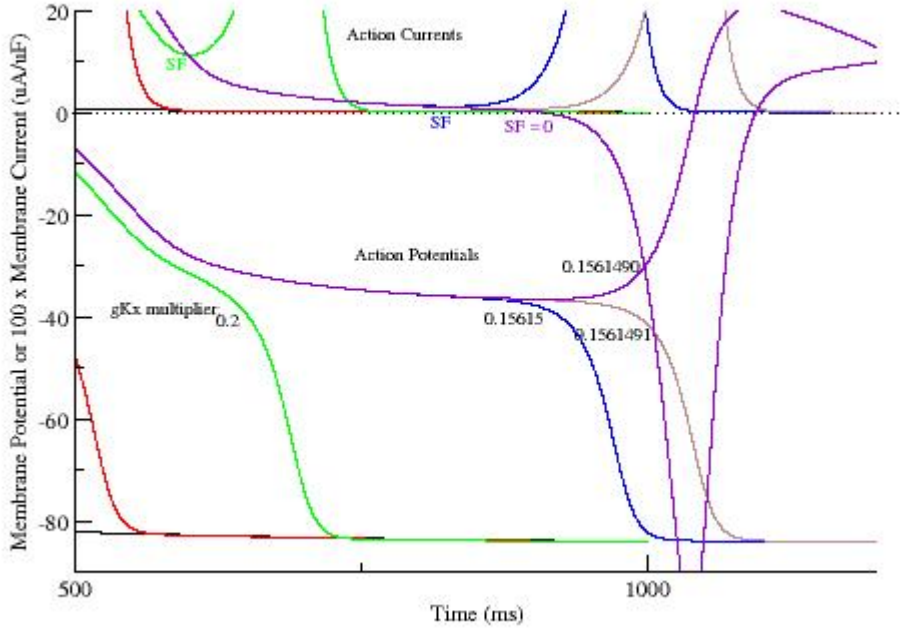
EAD spectra with BR (EADs were impossible with original parameters so gCa was set to 1.5 original value in order to force the resistance transition region to cross i=0 with slowed repolarization)



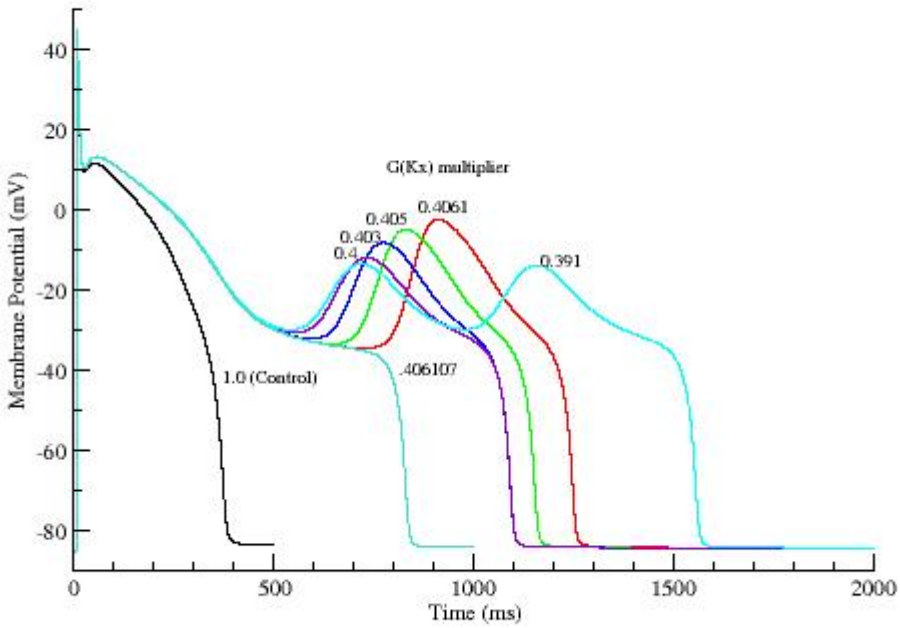
BR APs and associated action currents show a reduction in amplitude (SF) as repolarization is slowed.



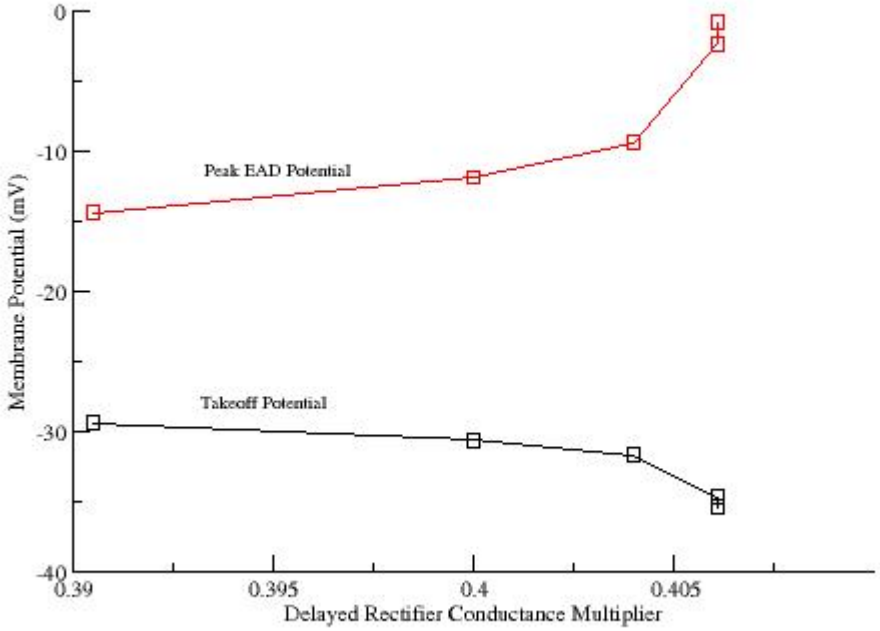
BR APs and currents with expanded axis reveals the coincidence of Vm with the minimum of the negative resistance region is zero.



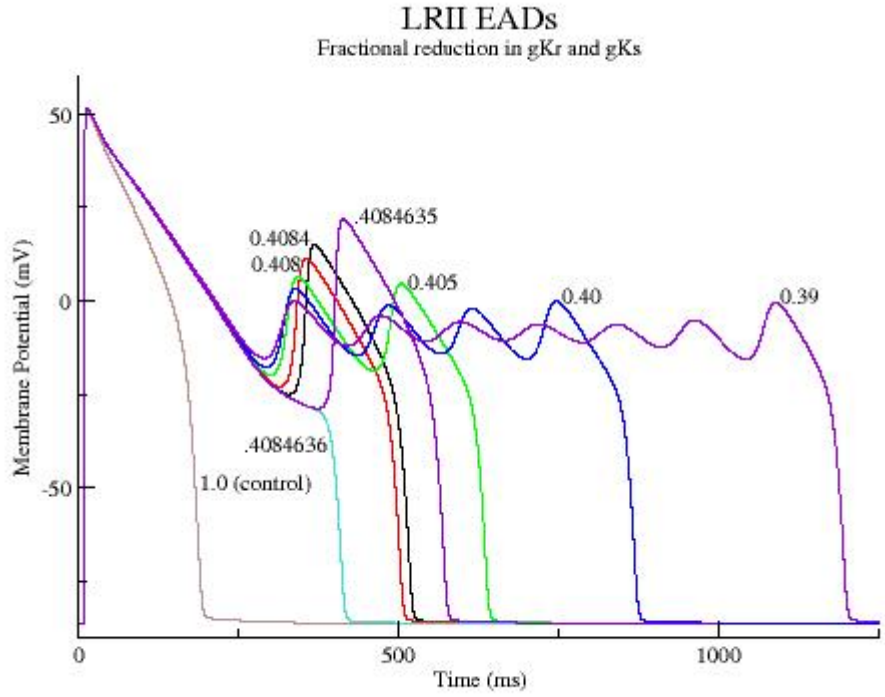
EAD spectra with LR 1



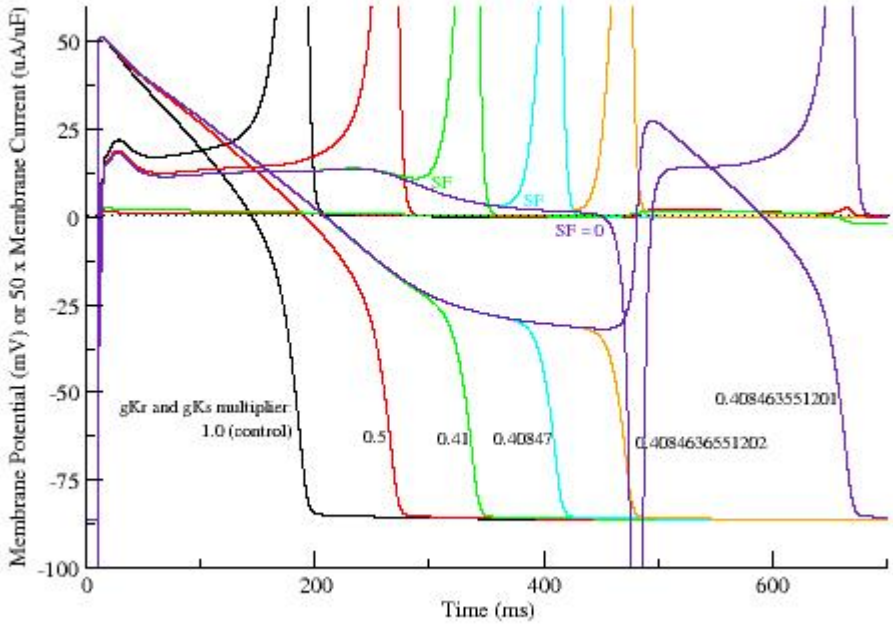
Plot of takeoff potentials and peak potentials for EADs generated by the LR 1 model.



EAD spectra with LRd (LR 2)



LRd APs and associated action currents reveal depolarizing shift in the minimum of the negative resistance region and reduction in amplitude (SF) as depolarization is slowed.



Theoretical considerations

The temporal trajectory of the repolarizing membrane potential (Vm) is dictated by the instantaneous current-voltage (i/v) which is a mixture of inward and outward components: C dVm/dt = -(I(in) + I(out)) . (Note that analysis of EAD mechanisms can be limited to the study of two generic currents - one inward and one outward. This is a useful way to explore the role of complexity in cardiac models and whether additional complexities alter the generic properites of the 2 current model). Depending on the nature of the individual i/v components, there may be regions of positive resistance (where increases in potential result in increases in current) or negative resistance (where increases in potential result in decreases in current). As the AP repolarizes, some outward currents deactivate while some inward current carriers reactivate. As seen with the above equation, when inward current dominates, dV/dt > 0 (depolarization) while when outward currents dominate, dV/dt < 0 (repolarization).

As the membrane potential follows the repolarization trajectory, what is the relationship between the repolarizing potential and the evolving i/v and specifically the growing source of inward current? During repolarization, nothing unusual happens as long as the negative resistance region falls above i=0. When Vm reaches an outward current negative resistance region, repolarization is accelerated. If, during repolarization, part of the nomally outward negative resistance region evolves such that it crosses the i=0 axis and becomes inward, then EADs are possible.

From numerical studies, the amplitude of the minimum of the negative resistance region of the repolarizing i/v curve (Safety Factory, SF) accurately measures EAD propensity. The SF can be measured under equilibrium conditions established at the rest potential (which is equivalent to a long pause after a pulse train). A SF profile can be measured as a function of delay following a train of conditioning pulses. The impact of altered inward or outward currents on EAD propensity are readily available by considering the potential alteration of the SF profile. Drugs and channel mutations that decrease the amplitude of the SF profile would be considered arrhythmogenic.

Delayed afterdepolarizations

Concept: they are oscillations of the membrane potential that occur after having completed phase 3 of AP or in phase 4. When they reach the limit, they trigger a new AP. They are observed in high rates (tachycardia-dependent). Their mechanism is caused by the opening of the INS. channel, sensitive to intracellular Ca2+ concentration.

Delayed afterdepolarizations (DADs), on the other hand, begin during phase 4 - after repolarization is completed, but before another AP would normally occur. They are due to elevated cytosolic calcium concentrations, as might be seen with digoxin toxicity. (25;26) The overload of the sarcoplasmic reticulum may cause spontaneous Ca2+ release during repolarization, causing the released Ca2+ to exit the cell through the 3Na+/Ca2+-exchanger which results in a net depolarizing current.

CLINICAL CAUSES OF DADs

1) A second phase of Ischemia and reperfusion: Coronary occlusion leading to nearly total absence of myocardial perfusion is the major cause of lethal ischemic arrhythmia in humans. In this setting, intracellular acidosis rapidly develops and leads to accelerated K+ efflux from the myocyte. Other metabolites, including lipid amphiphiles such as LPC, also rapidly accumulate in the ischemic zone. Elevated extracellular K+ and LPC cause membrane depolarization, which leads to slow conduction and increased refractoriness. These electrophysiologic changes contribute to the development of re-entrant rhythms, which predominate during early ischemia (phase 1a). Diffusion of extracellular K+ from the ischemic zone and release of endogenous catecholamines result in improvement in electrophysiologic parameters and are associated with a short arrhythmia-free interval, which occurs approximately 10 minutes after coronary occlusion. A second phase of arrhythmia (1b) then occurs and may be due in part to catecholamine-mediated triggered activity. Irreversible cell injury occurs 15 to 20 minutes after coronary occlusion and is associated with cell Ca++

overload, loss of gap junctions, and impaired cell coupling. This may lead to re-entrant arrhythmias. Reperfusion of ischemic myocardium leads to arrhythmia predominantly mediated by non re-entrant mechanisms. In humans, these reperfusion arrhythmias are usually relatively benign.(27).

2) Digitalis intoxication: atrial, junctional, fascicular and ventricular tachycardia(28).

3) Adrenergic stress on edocardium catecholamine-dependent VT Under conditions of enhanced calcium entry, myocytes closer to the endocardium exhibit a higher level of diastolic calcium and greater ectopic activity compared to the epicardium. Was observed simultaneous DAD and spontaneous calcium release events from myocytes in a normally coupled multicellular preparation. Myocytes near the endocardium are more susceptible to calcium-mediated triggered activity. (29).

4) Hypercalcemia consequence of enhanced sympathetic tone(30).

5) Multifocal or chaotic atrial tachycardia: multiple foci with triggered automaticity by delayed after potentials in phase 4, originated by: increase of circulating catecholamines, hypoxia, increase of CO2, hypopotassemia, hypomagnesemia, etc.

6) Idiopathic VT of the right and left ventricular outflow tract (RVOT and LVOT) and some idiopathic VT

7) Catecolaminergic Polymorphic Ventricular Tachycardia.

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